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Direct and indirect effects of diesel fuel on microphytobenthos and meiofauna in saltmarsh sediments

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Resultsprovide several insights into the influence of diesel contaminants on benthic food webs. Benthic microalgae constitute the base of this food web. Ths consistency of results from 1994 and 1995 experiments indicates the robustness of our observations of reduced copepod grazing and enhanced algal biomass in the presence of high levels of diesel contamination. While the broad responses of the two communities were similar, differences in the response of the meiofaunal communities are consistent with the hypothesis that the MS community is more sensitive to contaminants. Microalgal biomass in sediments is controlled primarily by meiofaunal grazing pressure. Mortality to meiofaunal grazers releases algae from grazing pressure. The algae grow until they become N-limited. Enhanced ammonium production in diesel-contaminated sediments then allows algal blooms to develop further. Thus, our experiments have allowed us to identify the general process by which blooms in diesel-contaminated occur.

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FINAL REPORT

GRANT #: 96PR01926-00

PRINCIPAL INVESTIGATOR: Thomas S. Bianchi

INSTITUTION: Tulane University

GRANT TITLE: Direct and indirect effects of diesel fuel on microphytobenthos and meiofauna in saltmarsh sediments

AWARD PERIOD: - 30 Oct. 1998

OBJECTIVES: To determine if(i) algal blooms in contaminated sediments are the result of direct or indirect stimulation from diesel or from a reduction in meiofaunal grazing; (ii) mortality to copepods in contaminated sediments is from direct toxicity from diesel, anoxic conditions, or a combination of the two; and (iii) *Cletocamptus deitersi* (a meiofaunal copepod) thrives in contaminated sediments because of reduced competition from other species, or exploitation of altered algal resources.

APPROACH: Microcosms experiments were performed to determine the effects of diesel-contaminated sediment on microalgal-meiofaunal interactions. Natural sediments were defaunated, then manipulated to control meiofaunal abundance and species composition, as well as diesel contamination. Treatments were compared to determine if reduced grazing or altered nutrient availability in diesel-contaminated sediments lead to algal blooms. Toxicity tests were used to determine interspecific differences in sensitivity to diesel and oxygen depletion. The influence of diesel on meiofaunal competitive interactions (grazing rates) was measured in communities manipulated to mimic those that occur in diesel-contaminated sediments. Microcosm experiments were performed to determine if enhanced N availability in diesel-contaminated sediments was from decaying carcasses of meiofauna, or altered sedimentary biogeochemical processes. HPLC analysis was employed to determine changes in sedimentary microphytobenthos, and how those changes relate to copepod feeding.

ACCOMPLISHMENTS: The effects of altered grazing and nutrient availability were examined in two microcosm experiments. In the first, we determined if diesel contamination influences NH_4^+ flux from sediments. Results indicate an enhanced flux of NH_4^+ from contaminated sediments. NH_4^+ concentrations remained very low in contaminated sediments incubated in the light, because microalgae consumed it as fast as it was produced. Addition of NH_4^+ to uncontaminated sediments did not stimulate algal growth, however, indicating that N availability was not the primary factor limiting algal growth.

In a second experiment, we examined the hypothesis that reduced grazing led to the formation of algal blooms. Meiofaunal abundance was manipulated to remove copepods and other larger grazers from sediment. Algal responses to this manipulation of grazing were compared to those that occurred in diesel-contaminated sediments. Both removal of grazers and contamination with diesel produced a 2-fold increase in algal biomass over a 4 days. Microscopic and HPLC analyses indicate that the increased algal biomass was almost entirely from growth of diatoms.

Experiments were also conducted to distinguish between the effects of hypoxia vs direct toxicity of diesel on meiofaunal copepods. In all species examined, we observed that hypoxia greatly increased the toxicity of diesel-contaminated sediment. *Cletocamptus deitersi*, a species that seems to be resistant to diesel toxicity, was the least sensitive of the species tested.

In previous microcosm experiments using natural communities, we observed that nematodes and *C. deitersi* thrive and exhibit enhanced grazing rates on microphytobenthos, leading to the hypothesis that *C. deitersi* and nematodes benefit from a release from competition for food when other copepods are killed. This hypothesis was tested by experimentally manipulating *C. deitersi* and nematode abundances to simulate conditions that occur in diesel-contaminated sediment. Grazing by *C. deitersi* and nematodes increased in the absence of copepod competitors. Among nematodes, enhanced grazing on algae was limited to a diatom feeder, *Ptycholaimellus* sp. Our evidence indicates that increased grazing by *C. deitersi* in contaminated sediments is partially from reduced competition, and partially a response to the enhanced algal biomass in contaminated sediments.

Our research indicates that algal blooms in diesel-contaminated sediments are a consequence of reduced grazing (from meiofaunal mortality) over short periods (5-14 days), followed by a second growth phase that is fueled by enhanced N fluxes. We performed two experiments to examine the source of this excess nitrogen. The first examined the possibility that N comes from decaying carcasses of animals killed via diesel toxicity. Results indicate that nitrogen released from decaying carcasses occurs over very short periods (3-4 days), well before algae become N-limited. Thus, excess N that fuels growth of algae must come from other N pools in the sediment.

Observations above led to an experiment (performed last fall) in which we examined in more detail the biogeochemistry of N dynamics in diesel-contaminated sediment. We are in the final stages of analyzing samples from this experiment. We are examining the hypothesis that altered microbial activity in diesel-contaminated sediment may result in increased metabolism of refractory marsh detritus, which contains geoplymers that may have "complexed" DIN; this could result in the release of organic and inorganic nitrogen. We are measuring total hydrolyzable and dissolved free amino acids (THAA) in experimental sediments from diesel-contaminated and control sediments, as well as N and P dynamics in inorganic pools (PO4, NH4, NO3, and NO2), and bulk sediment nitrogen. Analysis of inorganic pools has been completed, and analysis of amino acids and bulk sediment is underway. Preliminary analysis indicates that diesel contamination dramatically enhances the NH_4 (as previously documented) and PO_4 flux from contaminated sediments relative to controls. Evidence of enhanced PO_4 flux is new, and may contribute to the formation of microphytobenthos blooms in contaminated sediments. When amino-acid and bulk-sediment analyses are completed, we should have a much clearer picture of the origin of excess nitrogen that fluxes from contaminated sediments.

CONCLUSIONS: Results provide several insights into the influence of diesel contaminants on benthic food webs. Benthic microalgae constitute the base of this food web, and thus their dramatic increase in response to contaminants is ecologically significant. The consistency of results from 1994 and 1995 experiments indicates the robustness of our observations of reduced copepod grazing and enhanced algal biomass in the presence of high levels of diesel contamination. While the broad responses of the two communities were similar, differences in the response of the meiofaunal communities are consistent with the hypothesis that the MS community is more sensitive to contaminants. Microalgal biomass in sediments is controlled primarily by meiofaunal grazing pressure. Mortality to meiofaunal grazers (which occurs as a consequence of the interactive effects of hypoxia and diesel toxicity) releases algae from grazing pressure. The algae grow until they become N-limited. Enhanced NH_4^+ production in diesel-contaminated sediments then allows algal blooms to develop further. Thus, our experiments have allowed us to identify the general process by which blooms occur in diesel-contaminated sediments.

SIGNIFICANCE: Collectively, our research has yielded an understanding of how and why benthic communities in salt marshes respond to diesel. Diesel contamination dramatically alters benthic invertebrate community structure, grazing on microphytobenthos, and nitrogen biogeochemistry. These cascading effects lead to algal blooms and fundamental changes in competitive interactions among benthic invertebrates.

AWARD INFORMATION: T. Bianchi was awarded tenure in 1998.

PUBLICATIONS AND ABSTRACTS

Bennett, A., T.S., Bianchi, J.C. Means, & K. Carman. The effects of PAH contamination and grazing on the abundance and composition of microphytobenthos in salt marsh sediments (Pass Fourchon, LA): I. A microcosm experiment . J. Exp. Mar. Biol. Ecol.(Submitted; in revision)

Carman, K.R., Fleeger, J.W., & S.M. Pomarico. Does chronic hydrocarbon contamination alter the response of benthic communities to diesel contamination? Mar. Environ. Res. (Submitted, in revision).

Bennett, A., T.S. Bianchi, & J. C. Means. The effects of PAH contamination and grazing on the abundance and composition of microphytobenthos in salt marsh sediments (Pass Fourchon, LA):II. A field study. Estuar. Coastal Shelf Sci. (Submitted).

Carman, K.R. & J.C. Means. Sediment bacterial response to hydrocarbon contamination in two coastal saltmarshes. Estuaries. (In prep)

Carman, K.C., Bianchi, T.S., & J.W. Fleeger. Microalgal blooms in hydrocarbon-contaminated saltmarsh sediments: The influences of grazing and nitrogen availability. Mar. Ecol.

Prog. Ser. (In prep)

Bianchi, T.S. & K.R. Carman. The effects of PAH contamination on the availability of nitrogen to microphytobenthos in marsh sediments Org. Geochem. (In prep).

Abstracts

1998 *Benthic Ecology Meeting* (4)

Invited seminars given by Carman at Florida State University, Old Dominion Univ., Inst. for Marine Sciences (UNC Chapel Hill marine lab), Univ. North Carolina Chapel Hill, Univ. South Carolina

1997 *American Society for Limnology & Oceanography* (3)